



# First report of *Dendrorchis retrobiloba* Volonterio & Ponce de León, 2005 (Digenea, Gorgoderidae) in *Astyanax* aff. *fasciatus* (Cuvier, 1819) (Characiformes, Characidae) from southern Brazil

Moisés Gallas, Laura Roberta Pinto Utz

Laboratório de Ecologia Aquática, Escola de Ciências, Pontifícia Universidade Católica do Rio Grande do Sul, Avenida Ipiranga, 6681, Partenon, Porto Alegre, Rio Grande do Sul, 90619-900, Brazil.

**Corresponding author:** Moisés Gallas, [mgallas88@gmail.com](mailto:mgallas88@gmail.com)

## Abstract

*Dendrorchis retrobiloba* Volonterio & Ponce de León, 2005 was first described from the swim bladder of *Astyanax fasciatus* (Cuvier, 1819) from Montevideo, Uruguay. In this study, we necropsied specimens of *A. aff. fasciatus* collected from Lake Guaíba, in Rio Grande do Sul, Brazil, for the analysis of their helminths. The digeneans were identified as *D. retrobiloba* by their elongated body, oral sucker longer than ventral sucker, and 2 posterior lobes. This is the first report of *D. retrobiloba* in *A. aff. fasciatus* from Lake Guaíba and extends the known geographic distribution of this parasite.

## Key words

Characid, digenean, endoparasite, Neotropical Region, trematode.

**Academic editor:** Fernando Carbayo | Received 17 January 2019 | Accepted 23 April 2019 | Published 10 May 2019

**Citation:** Gallas M, Utz LRP (2019) First report of *Dendrorchis retrobiloba* Volonterio & Ponce de León, 2005 (Digenea, Gorgoderidae) in *Astyanax* aff. *fasciatus* (Cuvier, 1819) (Characiformes, Characidae) from southern Brazil. Check List 15 (3): 357–361. <https://doi.org/10.15560/15.3.357>

## Introduction

The family Gorgoderidae Looss, 1899 encompasses 3 subfamilies, Anaporrhutinae Looss, 1901, Gorgoderinae Looss, 1899, and Degeneriinae Cutmore, Miller, Curran, Bennett & Cribb, 2013 (Campbell 2008, Cutmore et al. 2013). The Gorgoderinae includes digenetic trematodes with intercaecal testes, the pharynx absent or present, and the seminal receptacle and cirrus sac absent (Campbell 2008). These parasites infect primarily the urinary system of freshwater and marine fishes but are also found in amphibians.

The genus *Dendrorchis* was proposed by Travassos (1926) to accommodate *Dendrorchis neivai* Travassos, 1926, a parasite of the swim bladder of *Brycon lundii*

(= *Brycon orthotaenia* Günther, 1864) from São Paulo state, Brazil. Subsequently, several other *Dendrorchis* species have been described in fish hosts, including *Dendrorchis retrobiloba* Volonterio & Ponce de León, 2005 in *Astyanax fasciatus* (Cuvier, 1819) from Montevideo, Uruguay (Volonterio and Ponce de León 2005), *Dendrorchis ritata* Soofi, Birmani & Dharejo, 2016 in *Rita rita* (Hamilton, 1822) from Jamshoro, Pakistan (Soofi et al. 2016), and *Dendrorchis pampae* Monteiro, Wendt & Zebral, 2018 in *Cynopoecilus melanotaenia* (Regan, 1912) from Rio Grande do Sul state, Brazil (Monteiro et al. 2018).

*Astyanax* Baird & Girard, 1854 is a relatively diverse characid genus, with 146 valid species (Froese and Pauly 2018) from a number of different geographic

regions. Four *Astyanax* species—*Astyanax eigenmanniorum* (Cope, 1894), *Astyanax* aff. *fasciatus* (Cuvier, 1819), *Astyanax henseli* Melo & Buckup, 2006, and *Astyanax lacustris* (Lütken, 1875)—are known to occur in Lake Guaíba, Rio Grande do Sul (Lucena et al. 2013). The helminth fauna of these species is still very poorly known. Until now, 2 species of digenetic trematodes, 2 monogeneans, and 1 nematode have been recorded in *Astyanax* from Lake Guaíba (Fortes and Hoffmann 1999, Gallas et al. 2014, 2015, 2016). In the present study, we provide the first report of *D. retrobiloba* in *A. aff. fasciatus* from Lake Guaíba, which extends this parasite's known geographic distribution in the Neotropical Region to southern Brazil.

## Methods

Specimens of *A. eigenmanniorum* ( $n = 20$ ), *A. aff. fasciatus* ( $n = 46$ ), *A. henseli* ( $n = 5$ ), and *A. lacustris* ( $n = 25$ ) from Lake Guaíba were examined for the presence of digenetic trematodes. The fish were collected with seine nets between January 2017 and October 2017 at Pintada Island, on Lake Guaíba, in the municipality of Porto Alegre, Rio Grande do Sul state, Brazil. All specimens were frozen until necropsy. The endoparasites found during the examination were fixed in A.F.A. for 48 hours (Gallas et al. 2017) and then stored in 70° GL ethanol before being stained in Delafield's hematoxylin. The specimens were clarified in cedarwood oil and mounted in Canada balsam for morphological identification. Digenean systematics follows Gibson et al. (2002) and Bray et al. (2008).

Unless otherwise indicated, measurements of the adult specimens ( $n = 3$ ) are given in micrometers ( $\mu\text{m}$ ), with the range of values for a given parameter being followed by the mean and the standard deviation ( $\pm$ ). In the case of the eggs, the number of eggs measured is cited. The terms forebody and hindbody follow Yamaguti (1971). Line drawings of the specimens were obtained using a Zeiss Axiostar microscope, with CorelDraw X4® and Adobe Photoshop® CS2. The infection parameters, i.e., prevalence and the intensity and amplitude of infection, follow Bush et al. (1997). A voucher specimen of the helminth was deposited in the Helminthological Collection of the Natural Sciences Museum (Coleção Helminológica do Museu de Ciências Naturais, CHMU) at the Lutheran University of Brazil in Canoas, Rio Grande do Sul, Brazil. The hosts were identified following Bertaco and Lucena (2010) and Lucena et al. (2013).

## Results

Phylum Platyhelminthes Gegenbaur, 1859  
Class Trematoda Rudolphi, 1808  
Subclass Digenea Carus, 1863  
Superfamily Gorgoderioidea Looss, 1899  
Family Gorgoderidae Looss, 1899  
Subfamily Gorgoderinae Looss, 1899

Genus *Dendrorchis* Travassos, 1926

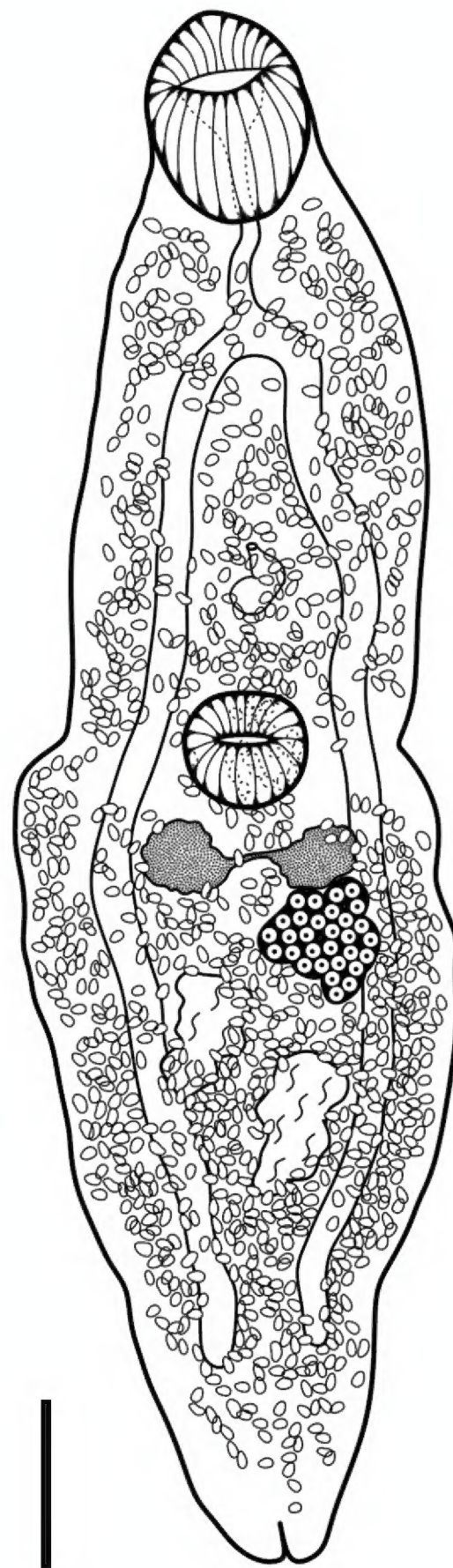
### *Dendrorchis retrobiloba* Volonterio & Ponce de León, 2005

Figure 1

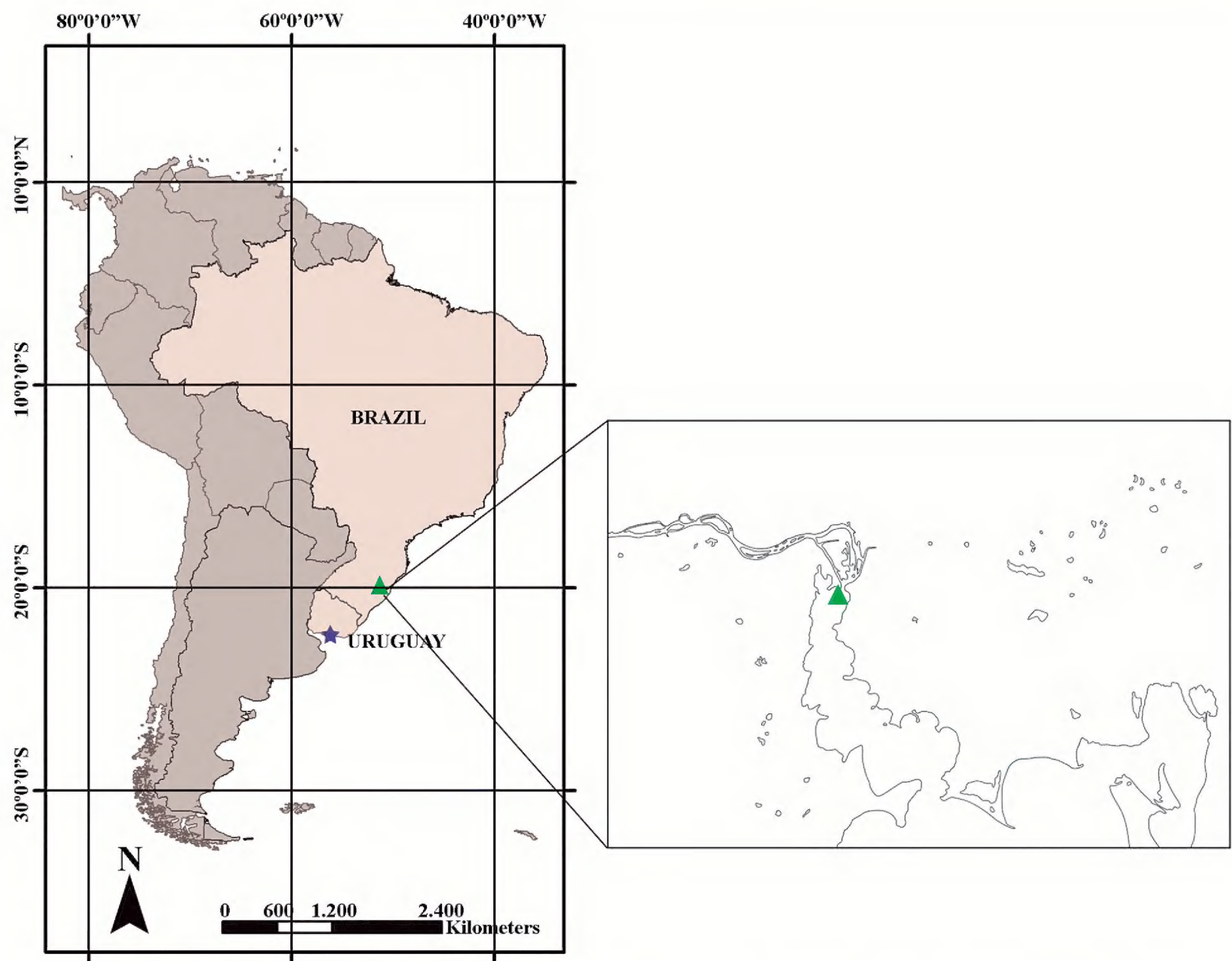
**Infection parameters.** Prevalence: 6.52%, mean intensity of infection: 1 helminth/host and mean abundance of infection: 0.06 helminth/host.

**New records.** Brazil: Rio Grande do Sul state: Municipality of Porto Alegre (30°17'11"S, 051°18'01" W) (Fig. 2). Host: *Astyanax* aff. *fasciatus* (Cuvier, 1819). Site of infection: swim bladder. Collection of parasites: 22 February 2018; identification of parasites: 5 June 2018. Voucher specimen of helminth deposited: CHMU-290-1-1.

**Identification.** This description is based on 3 stained specimens mounted in Canada balsam. Body elongated, 1.9–2.35 mm ( $2.17 \pm 0.25$  mm) long, with relatively narrow forebody, 0.42–0.51 mm ( $0.46 \pm 0.04$  mm) wide, in



**Figure 1.** *Dendrorchis retrobiloba* Volonterio & Ponce de León, 2005. Ventral view of whole specimen. Scale bar = 200  $\mu\text{m}$ .



**Figure 2.** Map showing Cañada Del Dragón, in the Departamento de Montevideo, Uruguay (blue star) the type locality of *Dendrorchis retrobiloba* Volonterio & Ponce de León, 2005, and the new locality, Lake Guaíba, in the state of Rio Grande do Sul, Brazil (green triangle).

comparison with the hindbody, 0.64–0.66 mm ( $0.65 \pm 0.01$  mm) wide. Oral sucker subterminal, 250–310 ( $280 \pm 30$ ) long, 220–270 ( $240 \pm 26$ ) wide. Ventral sucker smaller than oral sucker, 150–180 ( $167 \pm 15$ ) long, 160–170 ( $163 \pm 6$ ) wide, and 0.81–1.1 mm ( $0.97 \pm 0.14$  mm) from anterior extremity. Total body length to oral sucker ratio: 1:7.7; total body length to ventral sucker ratio: 1:13; oral sucker to ventral sucker ratio: 1:1.7. Pharynx absent. Esophagus 90–120 ( $107 \pm 15$ ) long, with caecal bifurcation closer to oral sucker than ventral sucker. Caecal bifurcation 330–440 ( $397 \pm 59$ ) from anterior extremity. Intestinal caeca terminating close to posterior extremity of body.

Genital pore in the forebody, 0.6–0.73 mm ( $0.67 \pm 0.07$  mm) from anterior extremity. No cirrus sac or cirrus observed. Seminal vesicle present, 65–90 ( $75 \pm 13$ ) long and 50–55 ( $52 \pm 3$ ) wide. Testes lobed and irregular in shape, arranged in diagonal, intercaecal, anterior testis 110–140 ( $127 \pm 15$ ) long, 70–80 ( $77 \pm 6$ ) wide; posterior testis 100–170 ( $147 \pm 40$ ) long, 70–100 ( $90 \pm 17$ ) wide. Seminal receptacle not observed. Ovary lobed, sinistral, intercaecal, posterior to vitellaria, pretesticular, 170–190 ( $180 \pm 10$ ) long, 170 wide. Mehli's gland and Laurer's canal not observed. Vitellaria composed of 2 lobed masses, located between ventral sucker and ovary. Right vitellarium 100–120 ( $113 \pm 11$ ) long, 110 wide; left

vitellarium 100–120 ( $110 \pm 10$ ) long, 80–130 ( $107 \pm 25$ ) wide. Uterus inter- and extra-caecal, in forebody and hindbody. Eggs oval, 22–32 ( $28 \pm 3$ ;  $n = 10$ ) long, 17–20 ( $19 \pm 1$ ;  $n = 10$ ) wide. Posterior extremity with 2 lobes, and excretory pore between them.

The specimens of *D. retrobiloba* are easily distinguished from the congeners *D. neivai*, *D. ritata*, and *D. pampae* based on morphological traits such as body shape, the size and shape of the suckers, testes, ovary and, the extension of the uterus (Travassos 1926, Volonterio and Ponce de León 2005, Soofi et al. 2016, Monteiro et al. 2018). The bodies of *D. neivai*, *D. ritata*, and *D. pampae* are pyriform, whereas *D. retrobiloba* has an elongated body. The oral sucker of *D. retrobiloba* is larger than its ventral sucker, whereas in all the other species, these 2 suckers are of similar size. In addition, *D. retrobiloba* is the only *Dendrorchis* species that has 2 posterior lobes, with a posterior cleft (Volonterio and Ponce de León 2005).

The testes of *D. neivai* are larger than the lobed ovary, while in *D. ritata* the ovary is triangular and similar in size to the testes. Irregularly shaped testes are found in *D. pampae* and *D. retrobiloba*, but in *D. retrobiloba*, the lobed ovary is a little longer than the testes. Except for *D. neivai*, which has a more extracaecal uterus with few eggs

in the intercaecal area, all the other species have a uterus in the inter- or extra-caecal fields. The vitellarium in the specimens examined by Volonterio and Ponce de León (2005) was described as having two compact, pyriform masses, whereas the specimens examined in the present study presented vitellaria organized in 2 lobed masses.

## Discussion

The diagnosis of the genus *Dendrorchis* provided by Travassos (1926) was amended recently by Volonterio and Ponce de León (2005), Campbell (2008), and Monteiro et al. (2018). Travassos (1926) recognized specimens with a pyriform body, small cirrus sac, branched testes, deeply lobed ovary, and vitellaria occupying all body fields, except for the intercaecal field above the vitellaria. Volonterio and Ponce de León (2005) added the following diagnostic traits: body wider posterior to the ventral sucker, testes branched or with superficial lobes, seminal receptacle present or absent, vitellaria elongate or pyriform, uterus extending to anterior portion and posterior region of the body, eggs thin-shelled without opercula.

Brooks and MacDonald (1986) examined 2 syntypes of *D. neivai* deposited in the Helminthological Collection of the Oswaldo Cruz Institute and noted the absence of a cirrus sac, which was the principal reason for Campbell's (2008) proposal for a new amendment of the diagnosis of *Dendrorchis*. This amendment included the broad and foliated hindbody, cirrus sac and seminal receptacle absent, and testes branched. Monteiro et al. (2018) further amended the diagnosis to include the presence of a cirrus sac and modify the position of the genital pore.

Given these slightly different diagnoses of the genus *Dendrorchis* (Travassos 1926, Volonterio and Ponce de León 2005, Campbell 2008, Monteiro et al. 2018), it is necessary to consider the question of the cirrus sac. If the presence of cirrus sac is considered to be a characteristic of *Dendrorchis* species (Travassos 1926, Volonterio and Ponce de León 2005, Monteiro et al. 2018), *Dendrorchis* would be the only gorgoderine genus to feature this structure (Campbell 2008). All other gorgoderine genera (except *Phyllodistomoides* Brooks, 1977 [?] and *Xystretrum* Linton, 1910) lack a cirrus sac, but present a structure (seminal vesicle) that has been misidentified as a cirrus sac by some authors. In the genus *Xystretrum*, there is a pseudosinus sac, which forms a sac-like structure together with the terminal genitalia, while the metratrum does not unite with the male duct (Campbell 2008).

In the drawings by Travassos (1926), Volonterio and Ponce de León (2005), and Monteiro et al. (2018), there is no evidence of a cirrus portion in the interior of the structure, which these authors identified as the cirrus sac. The cirrus sac identified by these authors may in fact represent a seminal vesicle, a pseudocirrus sac, or a false cirrus sac, which comprises a sac containing the male terminal genitalia, but without any differentiation of the cirrus, as defined by Yamaguti (1971). There

are also considerable inconsistencies in the widths of the cirrus sac (30.7–42.9) and the internal seminal vesicle (20.4–63.0) of the specimens analyzed by Volonterio and Ponce de León (2005), which may have been the result of either an error of measurement or the misidentification of the structures.

The life cycle of the gorgoderids is still little known, and most of the studies have focused on species of the genus *Phyllodistomum* Braun, 1899 (Cribb 1987, Urabe et al. 2015). The first intermediate host of *Phyllodistomum* is typically a bivalve, with the emerging cercariae infecting arthropods (crustaceans or insects), other mollusks, or tadpoles, which act as the second intermediate host, with fish as the definitive host (Cribb 1987). The low prevalence of *D. retrobiloba* recorded in our study indicates that *A. aff. fasciatus* ingests small numbers of the intermediate hosts of *D. retrobiloba*, which is consistent with the composition of the contents of the stomachs and intestines of the fish specimens examined, which contained more plant matter than invertebrates. *Astyanax* species are considered to be opportunistic omnivores, ingesting the food items available in the environment (Vilella et al. 2002, Wolff et al. 2009). This may also account for the lack of *D. retrobiloba* in the other *Astyanax* species examined, given that *A. lacustris* is considered to be predominantly herbivorous, while *A. eigenmanniorum* and *A. henseli* consume mainly insects and plant matter (Vilella et al. 2002, Ferrari et al. 2015).

Prior to our study, only 2 species of digenetic trematodes (*Genarchella parva* Travassos, Artigas & Pereira, 1928 and *Zonocotyle bicaecata* Travassos, 1948) had been observed in *A. aff. fasciatus* from Lake Guaíba (Fortes and Hoffmann 1999). The new record represents the first report of *D. retrobiloba* in *A. aff. fasciatus* from southern Brazil and extends the known geographic distribution of *D. retrobiloba* from its only other known locality in Uruguay. The results of our study also provide important insights into the morphology of *D. retrobiloba* and other species of the genus.

## Acknowledgements

We thank the local fishermen for host collection and anonymous reviewers for their comments. MG thanks the Brazilian National Research Council (CNPq) for financial support (process number 140639/2016-0).

## Authors' Contributions

MG necropsied the hosts, prepared and identified the parasites, prepared figures, and co-wrote the text; LRP co-wrote the text.

## References

- Bertaco VA, Lucena CAS (2010) Redescription of *Astyanax obscurus* (Hensel, 1870) and *A. laticeps* (Cope, 1894) (Teleostei: Characidae): two valid freshwater species originally described from rivers of Southern Brazil. *Neotropical Ichthyology* 8 (1): 7–20.

- <https://doi.org/10.1590/S1679-62252010000100002>
- Bray RA, Gibson DI, Jones A (Eds) (2008) Keys to the Trematoda. Volume 3. CAB International, Wallingford, 824 pp.
- Brooks DR, MacDonald CA (1986) A new species of *Phyllodistomum* Braun, 1899 (Digenea: Gorgoderidae) in a Neotropical catfish, with discussion of the generic relationships of the Gorgoderidae. Canadian Journal of Zoology 64 (6): 1326–1330. <https://doi.org/10.1139/z86-197>
- Bush AO, Lafferty KD, Lotz JM, Shostak AW (1997) Parasitology meets ecology on its own terms: Margolis *et al.* revisited. Journal of Parasitology 83 (4): 575–583. <https://doi.org/10.2307/3284227>
- Campbell RA (2008) Family Gorgoderidae Looss, 1899. In: Bray RA, Gibson DI, Jones A (Eds) Keys to the Trematoda. Volume 3. CABI Publishing, Wallingford, 191–213.
- Cribb TH (1987) A new species of *Phyllodistomum* (Digenea: Gorgoderidae) from Australian and New Zealand freshwater fishes with notes on the taxonomy of *Phyllodistomum* Braun, 1899. Journal of Natural History 21 (6): 1525–1538. <https://doi.org/10.1080/00222938700770951>
- Cutmore SC, Miller TL, Curran SS, Bennett MB, Cribb TH (2013) Phylogenetic relationships of the Gorgoderidae (Platyhelminthes: Trematoda), including the proposal of a new subfamily (Dergeneriinae n. subfam.). Parasitology Research 112: 3063–3074. <https://doi.org/10.1007/s00436-013-3481-5>
- Ferrari L, Lampert VR, Azevedo MA (2015) Análise da dieta de *Astyanax henseli* (Characiformes, Characidae) do curso superior do Rio dos Sinos, RS, Brasil. In: XXI Encontro Brasileiro de Ictiologia, Recife.
- Froese R, Pauly D (Eds) (2018) FishBase. World Wide Web electronic publication, version (06/2018). <http://www.fishbase.org>. Accessed on: 2018-7-11.
- Fortes E, Hoffmann RP (1999) Trematódeos gastrintestinais de *Astyanax fasciatus* (Cuvier, 1819), lambaris do lago Guaíba, Porto Alegre, Rio Grande do Sul, Brasil. A Hora Veterinária 18 (108): 55–56.
- Gallas M, Calegari-Marques C, Amato SB (2014) A new species of *Cacatuocotyle* (Monogenea, Dactylogyridae) parasitizing two species of *Astyanax* (Ostariophysi, Characidae) in southern Brazil. Acta Parasitologica 59 (4): 638–642. <https://doi.org/10.2478/s11686-014-0283-6>
- Gallas M, Calegari-Marques C, Amato SB (2015) Supplemental observations on the morphology of *Procamallanus* (*Spirocamallanus*) *hilarii* (Nematoda: Camallanidae) parasitizing two species of *Astyanax* (Characiformes: Characidae) and ecological analyses. Revista Mexicana de Biodiversidad 86 (3): 590–596. <https://doi.org/10.1016/j.rmb.2015.07.005>
- Gallas M, Calegari-Marques C, Amato SB (2016) A new species of *Characithecium* (Monogenea: Dactylogyridae) from external surface and gills of two species of *Astyanax* (Ostariophysi: Characidae) in southern Brazil. Revista Mexicana de Biodiversidad 87 (3): 903–907. <https://doi.org/10.1016/j.rmb.2016.06.011>
- Gallas M, Silveira EF, Périco E (2017) *Cotylogaster basiri* Siddiqi & Cable, 1960 (Aspidobothrea: Aspidogastridae) in *Pogonias cromis* (Linnaeus, 1766) (Perciformes: Sciaenidae): first report from Brazil. Brazilian Journal of Biology 77 (1): 79–82. <https://doi.org/10.1590/1519-6984.10015>
- Gibson DI, Jones A, Bray RA (Eds) (2002) Keys to the Trematoda. Volume 1. CABI Publishing, Wallingford, 521 pp.
- Lucena CAS, Castro JB, Bertaco VA (2013) Three new species of *Astyanax* from drainages of southern Brazil (Characiformes: Characidae). Neotropical Ichthyology 11 (3): 537–552. <https://doi.org/10.1590/S1679-62252013000300007>
- Monteiro CM, Wendt EW, Zebral YD (2018) *Dendrorchis pampae* sp. n. (Digenea: Gorgoderidae) from *Cynopoecilus melanotaenia* (Cyprinodontiformes: Cynolebiidae) a killifish from southern Brazil, with an emended diagnosis of *Dendrorchis*. Journal of Helminthology: 1–5. <https://doi.org/10.1017/S0022149X18000172>
- Soofi H, Birmani NA, Dharejo AM (2016). *Dendrorchis ritata* n. sp. (Trematoda: Gorgoderidae) from catfish *Rita Rita* (Siluriformes: Bagridae) of Jamshoro district, Sindh, Pakistan. International Journal of Fauna and Biological Studies 3 (3): 17–19.
- Travassos L (1926) Trematodeos Novos. (V). Boletim biológico 1: 16–20.
- Urabe M, Ishibashi R, Uehara K (2015) The life cycle and molecular phylogeny of a gorgoderid trematode recorded from the mussel *Nodularia douglasiae* in the Yodo River, Japan. Parasitology International 64 (1): 26–32. <https://doi.org/10.1016/j.parint.2014.09.003>
- Vilella FS, Becker FG, Hartz SM (2002) Diet of *Astyanax* species (Teleostei, Characidae) in an Atlantic Forest River in southern Brazil. Brazilian Archives of Biology and Technology 45 (2): 223–232. <http://doi.org/10.1590/S1516-89132002000200015>
- Volonterio O, Ponce de León R (2005) Description of *Dendrorchis retrobiloba* n. sp. (Digenea: Gorgoderidae) from *Astyanax fasciatus* (Teleostei: Characidae) from southern Uruguay, with an emended diagnosis of the genus *Dendrorchis*. Journal of Parasitology 91 (5): 1204–1207. <https://doi.org/10.1645/GE-3465.1>
- Wolff LL, Abilhoa V, Rios FS, Donatti L (2009) Spatial, seasonal and ontogenetic variation in the diet of *Astyanax* aff. *fasciatus* (Ostariophysi: Characidae) in an Atlantic Forest river, southern Brazil. Neotropical Ichthyology 7 (2): 257–266. <http://doi.org/10.1590/S1679-62252009000200018>
- Yamaguti S (1971) Synopsis of digenetic trematodes of vertebrates. Volumes I and II. Keigaku Publishing, Tokyo, 1074 pp.